



Environmental Analysis Program

Pomona College
Biogeochemistry Laboratory

SOP-L408

Soil Respiration – Solvita CO₂ Burst Test

Standard Operating Procedure

SOP Number	SOP-L408
Revision	1.1
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Date	2026-06-26
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Table of contents

1 Purpose	2
2 Scope	2
3 Responsibilities	2
4 Definitions	2
5 Safety and Hazards	3
5.1 Required PPE	3
6 Equipment and Materials	3
7 Procedure	4
7.1 Sample Preparation	4
7.2 Incubation Setup	4
7.3 Incubation	4
7.4 Reading Results	4
7.5 Calculating Results	5
7.6 Cleanup	5
8 Quality Control	5
8.1 Routine QC	5
8.2 Corrective Actions	6
9 Data Interpretation	6
10 Waste Disposal	6
11 References	7
12 Related SOPs	7
13 Training Record	7

1. Purpose

This SOP describes the measurement of soil microbial respiration using the Solvita CO₂ burst test. The method quantifies CO₂ evolution from rewetted, air-dried soil as an index of microbial activity and labile carbon availability (Haney et al., 2008a; Haney et al., 2008b).

The CO₂ burst exploits the “Birch effect,” the well-documented pulse of microbial respiration that occurs when dry soil is rapidly rewetted (Birch, 1958; Fierer & Schimel, 2003). Drying and rewetting disrupts soil aggregates, lyses a fraction of the microbial biomass, and releases intracellular osmolytes, all of which provide readily mineralizable substrate that surviving microbes respire as CO₂ (Fierer & Schimel, 2002; Xiang et al., 2008). The magnitude of the burst integrates microbial biomass size, community physiological status, and the pool of labile organic carbon, making it a useful single-number indicator of biological soil fertility (Haney et al., 2008b).

2. Scope

- **Applies to:** Air-dried, sieved soil samples collected per [SOP-F400](#) for microbial activity assessment
- **Does not apply to:** Compost maturity testing (see [SOP-L413](#)); field-moist basal respiration; long-term (7–28 day) incubation-based respirometry; or chamber-based field CO₂ flux measurements (see [SOP-F406](#))
- **Location:** EA Program laboratory, SGM 133

3. Responsibilities

Role	Responsibility
Lab PI	Approve protocol, ensure training compliance
Lab Manager	Maintain Solvita kit inventory, verify probe expiration dates
Trained Personnel	Perform assay, record and report results
New Students	Complete training before independent use

4. Definitions

- **Basal respiration:** The rate of CO₂ production from soil under controlled, standardized conditions (specific moisture, temperature, incubation time) without additional substrate; reflects the size and activity of the soil microbial community.
- **CO₂ burst:** The pulse of CO₂ released from dry soil immediately upon rewetting; caused by osmotic stress on microorganisms, physical disruption of aggregates, and rapid oxidation of previously protected organic matter; the basis of the Solvita CO₂ Burst test.
- **Colorimetric gel pad:** The CO₂-sensitive indicator element in the Solvita kit; changes color in proportion to the CO₂ concentration in the jar headspace; read with a Solvita paddle reader or digitally.

- **Digital Color Reader (DCR):** The Solvita handheld device that reads the color of the gel pad and converts it to a numerical Solvita Index value.
- **Solvita Index:** A unitless scale (1–8) reported by the Solvita CO₂ Burst test; higher values indicate greater biological activity and organic matter availability; values correlate with potentially mineralizable nitrogen.
- **Soil microbial biomass:** The mass of living microbial cells (bacteria, fungi, protozoa) in soil; the primary driver of organic matter decomposition and nutrient cycling; estimated indirectly by CO₂ respiration or directly by fumigation-extraction (see SOP-L409).
- **Water-holding capacity (WHC):** The maximum amount of water a soil can hold after free drainage; expressed as a percentage of dry soil mass; the Solvita protocol adjusts sample moisture to a defined fraction of WHC for standardization.

5. Safety and Hazards

Hazards

- **Physical:** Minor risk of glass breakage from incubation jars. Handle jars carefully; clean up broken glass with a brush and dustpan, not bare hands.
- **Biological:** Soil samples may contain soil-borne pathogens. Wash hands thoroughly after handling soil. Do not touch your face during the procedure.

5.1. Required PPE

- ☐ Nitrile gloves
- ☐ Safety glasses
- ☐ Lab coat

6. Equipment and Materials

Item	Notes
Solvita CO ₂ burst test kit	Should include enough probes for all samples, duplicates and controls.
Solvita Digital Color Reader (DCR)	For quantifying gel color change; calibrate before each batch
Incubation jars (475 mL)	Glass jars with screw-top lids
30 cc soil scoop	Provided with kit
Small beakers	Can be plastic or glass. Must fit inside incubation jar
Analytical balance	Capable of 0.1 g resolution
10ml pipette + pipette pump	For adding deionized water
Deionized water	From Milli-Q system or equivalent
Incubator or controlled room	Set to 25 +/- 1 C
Timer	24-hour countdown
Lab notebook or data sheet	For recording results

7. Procedure

7.1. Sample Preparation

1. Use soil that has been collected per [SOP-F400](#).
2. If samples were stored at 4 C after sieving, allow them to equilibrate to room temperature (20–25 C) for at least 30 minutes before beginning the assay.
3. Sieve soils through a 2mm sieve. Use a mortar and pestle to break up soil clumps that did not pass through the sieve initially and resieve it. Discard any rocks or other materials that do not pass through the sieve.

Note

The CO₂ burst test is designed for air-dried soil. Drying standardizes the starting moisture across samples and maximizes the rewetting response. Results from field-moist soil are not comparable to published Solvita norms (Woods End Laboratories, 2019).

7.2. Incubation Setup

1. Measure 30ccs of the dried and sieved soils using a 30cc volume scoop and add it to a small beaker. Weight the soil (do not include weight of beaker) and record the exact mass to 0.1 g. Shake it gently to level the surface.
2. Gently sprinkle 9ccs of deionized water onto the surface of the soil using a pipette. The rewetting triggers the microbial CO₂ burst (Fierer & Schimel, 2003).
3. Remove a fresh CO₂-sensitive gel probe from its sealed foil packet. Inspect the gel: it should be attached to the probe, uniform in color with no cracks, discoloration, or signs of prior exposure. Discard compromised probes. Make sure not to touch the gel while handling the probe.
4. Insert the probe into the jar so that the gel face is exposed to the headspace above the soil surface. Do not bury the probe in soil.
5. Seal the jar immediately and securely.
6. Record the jar number, sample ID, soil mass, water volume added, and start time.

7.3. Incubation

1. Place sealed jars in the incubator set to 25 +/- 1 C
2. Incubate undisturbed for 24 hours
3. Do not open or move jars during incubation

7.4. Reading Results

1. After exactly 24 hours (+/- 15 minutes), remove the gel probe from the jar
2. Immediately insert the probe into the Solvita Digital Color Reader (DCR) and record the numerical reading

- Record the color number and the corresponding CO₂-C concentration (mg CO₂-C kg⁻¹ soil) from the DCR or the Solvita conversion table

 Tip

Read probes promptly after removing them from the jar. Prolonged exposure to ambient air will alter the gel color and compromise the reading. The manufacturer recommends reading within 2 minutes of opening the jar.

7.5. Calculating Results

Express results on a dry-soil basis:

$$\text{CO}_2\text{-C (mg kg}^{-1}\text{)} = \frac{\text{DCR reading (mg CO}_2\text{-C)}}{\text{oven-dry soil mass (kg)}}$$

If the oven-dry mass is not known, calculate it from the air-dried mass and the gravimetric moisture content of a companion subsample determined by [SOP-L400](#).

7.6. Cleanup

- Dispose of used soil in the designated waste container (return to field site if uncontaminated, or standard trash)
- Wash incubation jars thoroughly with soap and water; rinse with deionized water; dry completely before reuse
- Dispose of used gel probes in standard trash

8. Quality Control

8.1. Routine QC

QC Element	Requirement
Duplicates	At least 1 per 10 samples; duplicates should agree within 15% relative percent difference (RPD)
Control	One empty, sealed jar with gel probe per batch; probe should read at or near baseline
Incubator temperature	Verify and record at start and end of each 24 h period; must be 25 +/- 1 C
DCR calibration	Calibrate per manufacturer instructions before each batch
Gel probe storage	Store sealed in foil packets at room temperature, away from light; check expiration dates before use

8.2. Corrective Actions

- If the blank reading exceeds the baseline by more than one color increment, check for CO₂ contamination sources (e.g., proximity to gas burners, poor jar seals) and repeat the batch
- If duplicate RPD exceeds 15%, re-run the sample pair. Persistent poor precision may indicate sample heterogeneity; increase the number of subsamples

9. Data Interpretation

The CO₂ burst is proportional to microbial biomass and labile carbon and is correlated with potential carbon, nitrogen, and phosphorus mineralization (Haney et al., 2008b). Typical ranges for the Solvita CO₂ burst test on agricultural and wildland soils are given below, adapted from Haney et al. (2008a) and the Solvita technical literature (Woods End Laboratories, 2019):

CO ₂ -C (mg kg ⁻¹)	Interpretation	Implication
< 20	Very low	Severely depleted microbial activity; very low labile C
20–50	Low	Limited biological activity; management intervention may be needed
50–100	Moderate	Adequate microbial activity for many soils
100–150	High	Active microbial community; good labile C supply
> 150	Very high	Highly active soil; common in recently amended or high-organic soils

! Important

These thresholds are general guidelines. Interpret results in the context of soil type, land use, climate, and management history. Comparisons are most meaningful when made across treatments within a single study using the same sampling and analytical protocol.

10. Waste Disposal

Waste Type	Disposal Method
Used soil	Return to field site or standard trash
Used gel probes	Standard trash
Deionized water rinse	Drain
Incubation jars	Wash, rinse, and reuse

11. References

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- Xiang, S.-R., Doyle, A., Holden, P. A., & Schimel, J. P. (2008). Drying and rewetting effects on C and N mineralization and microbial activity in surface and subsurface California grassland soils. *Soil Biology and Biochemistry*, 40(9), 2281–2289. <https://doi.org/10.1016/j.soilbio.2008.05.004>

12. Related SOPs

SOP	Title	Relationship
F400	Soil Sampling and Processing	Sample collection and preparation
F406	Soil Respiration Chambers	Field respiration complement
L002	Laboratory Safety	Required safety training
L400	Soil Moisture (Gravimetric)	Moisture correction for dry-mass basis
L406	Loss on Ignition (SOM)	Complementary organic matter index
L409	Microbial Biomass Carbon (CFE)	Direct biomass measurement
L413	Solvita Compost Maturity	Related Solvita protocol for compost

13. Training Record

Before performing this assay independently, personnel must:

1. Read this SOP in full
2. Complete [SOP-L002 Laboratory Safety](#) training

3. Complete [SOP-F400 Soil Sampling](#) training (for sample preparation context)
4. Review the Solvita CO₂ Burst protocol sheet included in the kit
5. Observe a trained operator perform the full procedure (setup through DCR reading)
6. Perform the assay under supervision with acceptable QC results (duplicate RPD within 15%; blank within acceptable range)
7. Be signed off by the PI or lab manager